

Seed morphology of *Rhodocodon* (*Hyacinthaceae*) and its systematic implications

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with 10 figures and 1 table

Key words: *Hyacinthaceae*, *Rhodocodon*, *R. campanulatus*, *R. cryptopodus*, *R. cyathiformis*, *R. floribundus*, *R. giganteus* ined., *R. graciliscapus*, *R. intermedius*, *R. jackyi*, *R. mascarenensis*, *R. monophyllus*, *R. perrieri* ined., *R. rotundus*, *R. urGINEOIDES*. – Seeds, morphology, SEM.

Summary

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As part of studies of seeds in *Hyacinthaceae*, we detected the genus *Rhodocodon* as a rare example in the family that shows remarkable variability in seed morphology. Detailed descriptions and a key for the seeds of 13 species and their systematic implications are presented.

1. Introduction

The family *Hyacinthaceae* (sensu APG 2003) contains about 700–1000 species of bulbous plants distributed in Africa, Europe and Asia. With *Oziroë* (RAFINESQUE 1837: 53) *Hyacinthaceae* are also represented in South America (SPETA 1998a, b, APG 2003). In APG 2009 and APG 2016, *Hyacinthaceae* are treated as *Asparagaceae* subfamily *Scilloideae*, and the former accepted subfamilies *Hyacinthoideae*, *Ornithogaloideae*, *Oziroëoideae* and *Urgineoideae* (APG 2003, PFOSSE & SPETA 1999, MANNING & al. 2004, MARTÍNEZ-AZORÍN & al. 2011) are reduced to tribes. However, we favor *Hyacinthaceae* for morphological reasons.

The Malagasy genus *Rhodocodon* BAKER 1881: 280, as part of the subfamily *Urgineoideae*, forms a well-supported monophyletic clade. Further information on *Urgineoideae* or *Rhodocodon* and generic circumscriptions are given in SPETA 1998a, b, MARTÍNEZ-AZORÍN & al. 2013a, b and KNIRSCH & al. 2015, 2016. Recently, MANNING 2018 treated the genus *Rhodocodon* as *Drimia* sect. *Rhodocodon* (BAKER) J. C. MANNING & GOLDBLATT. However, based on evident divergences in morphology and molecular data with regards to other genera in *Urgineoideae*, we do

not follow this concept. According to the latest revision (KNIRSCH & al. 2015, 2016), the genus consists of 14 species.

In many plant descriptions, seeds are characterized only by a short note regarding their size and/or color. However, seed morphology is usually rich and diverse, especially in *Hyacinthaceae* (SPETA 1998a, b, BEDNORZ & CZARNA 2008). The work of BARTHLOTT 1981 on cell shape, curvatures and cell wall structures demonstrates the importance of these data for systematic studies.

Various works (WETSCHNIG & al. 2002, MARTÍNEZ-AZORÍN & al. 2006, 2007, 2009, KOLLER 2010, LUIDOLD 2010) demonstrate the wide morphological range of seeds in *Hyacinthaceae*.

Seeds of other genera of *Hyacinthaceae* often show specific and relatively homogeneous common features that allow genus discrimination. However, *Rhodocodon* is a rare example in the family that shows a remarkably high variability in seed morphology. For example, the seeds of *R. urGINEOIDES* with a thousand-seed weight (TSW) of 0.21 g are among the lightest in the family, whereas seeds of *R. giganteus* with a TSW of 26.43 g range among the heaviest ones. Light microscopic analyses, e.g. of seed and embryo shape, embryo curvature or testa

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color are useful parameters to distinguish six different seed types (Fig. 1A–F).

The aim of the present work is to describe the seed morphology of 13 *Rhodocodon* species (Table 1) in detail and to check their taxonomic suitability.

2. Material and methods

Seed samples (Table 1) were obtained from the living plant collection cultivated at the Botanical Garden of the former Institute of Plant Sciences (now Institute of Biology) of the University of Graz and from the private collection of W. KNIRSCH. All seed samples are stored in the *Hyacinthaceae* seed collection of W. WETSCHNIG.

Detailed morphological studies of *Rhodocodon* species were conducted on fresh or air-dried seeds, which were photographed (lateral and raphe view, longitudinal and cross section) using a digitalcam (Zeiss, AxioCam MRc) mounted on a light microscope (Zeiss, Stemi 2000C). The pictures were visualized and analyzed by the program AxioVision SE 64. We took several photographs at different focus distances and combined the stack with Helicon Focus Pro into a fully focused image.

Color terms are based on the RAL color system.

For the SEM investigations, dry seeds were mounted on specimen stubs using double-sided adhesive tape and sputtered with gold in an agar sputter-coater and scanned with a Philips XL30 ESEM at 20kV. The AxioVision SE 64 software was used also here to generate the data.

The values in the seed descriptions are mean values. We selected three representative seeds from one plant of every species (Table 1) to generate the data concerning embryo, endosperm and seed morphology. The number of seeds used for the calculation of the thousand-seed weight (TSW) is given in Table 1. Cell area and perimeter are average values of multiple measurements along the seed surface.

The morphological terminology follows BARTHOLOTT 1981, 1984, BARTHOLOTT & EHLE 1977 and WERKER 1997 and has been adapted and modified if necessary.

Seed descriptions and the key were generated with DELTA Editor (DALLWITZ 1980, DALLWITZ & al. 1999) and Intkey (DALLWITZ 1980, DALLWITZ & al. 1993, 1995, 2000).

3. Results

3.1. *Rhodocodon campanulatus* KNIRSCH, MART.-AZORÍN & WETSCHNIG (Fig. 1F, 2a–f)

Embryo: Shape of longitudinal section cylindrical, curved. Length of axis 1.87 mm. Width 0.57 mm. Height 0.68 mm. Area of longitudinal section 0.85 mm². Area of cross section 0.3 mm².

Table 1: Voucher data for specimens used in the studies

Specimen	Voucher ^a	Voucher ^b	TSW ^c
<i>R. campanulatus</i>	04582	02446-02	9
<i>R. cryptopodus</i>	04686	02416-06	20
<i>R. cyathiformis</i>	04679	04479-05	18
<i>R. floribundus</i>	04683	04478-05	19
<i>R. giganteus ined.</i>	03843	02514-01	7
<i>R. graciliscapus</i>	05025	04477-07	6
<i>R. intermedius</i>	04682	04475-05	14
<i>R. jackyi</i>	04684	04669-06	60
<i>R. mascarenensis</i>	03708	–	38
<i>R. monophyllus</i>	04778	04677-01	4
<i>R. perrieri ined.</i>	04774	04567-01	23
<i>R. rotundus</i>	04586	02407-01	13
<i>R. urGINEOIDES</i>	03674	02545-02	–
<i>R. urGINEOIDES</i>	02432	–	34

^a) Seed

^b) Plant

^c) Number of seeds available for TSW calculation

Endosperm: Not oblate, D-shaped in longitudinal section, circular in cross section, with smooth outline. Length 2.75 mm. Width 2.31 mm. Height 2.34 mm. Area of longitudinal section 4.48 mm². Area of cross section 4.45 mm².

Seed: D-shaped in lateral view and in cross section. Shape approximately corresponding to the shape of the endosperm, defined by endosperm and testa. Thousand-seed weight 8.1 g. Length 4.13 mm. Height 3.9 mm. Width 2.92 mm. Area of lateral view 10.66 mm². Area of longitudinal section 9.8 mm². Area of cross section 8.53 mm². Edges/strips/wings absent. Testa not connected with the endosperm, without appendices, black, glossy, cell pattern visible. Raphe with different color than testa. Hilum colored like testa.

Seedcoat surface: Raphe elevated. Stomata absent.

Primary sculpture: Cell area 10590 µm². Cell perimeter 596.94 µm. Frame width 174.15 µm. Frame height 171.4 µm. Cell shape uniform, generally elongate-polygonal. Anticlinal cell walls irregularly lobate in surface view, boundaries channeled, cell corners not divergent, periclinal cell wall plane.

Secondary sculpture: Surface of the outer cell wall foveolate, with waxes.

Tertiary sculpture: Wax layer plane, smooth.

3.2. *Rhodocodon cryptopodus* (BAKER) KNIRSCH, MART.-AZORÍN & WETSCHNIG (Fig. 1F, 2g–l)

Embryo: Shape of longitudinal section cylindrical, straight. Length of axis 1.06 mm. Width 0.15 mm. Height 0.35 mm. Area of longitudinal section 0.22 mm². Area of cross section 0.04 mm².

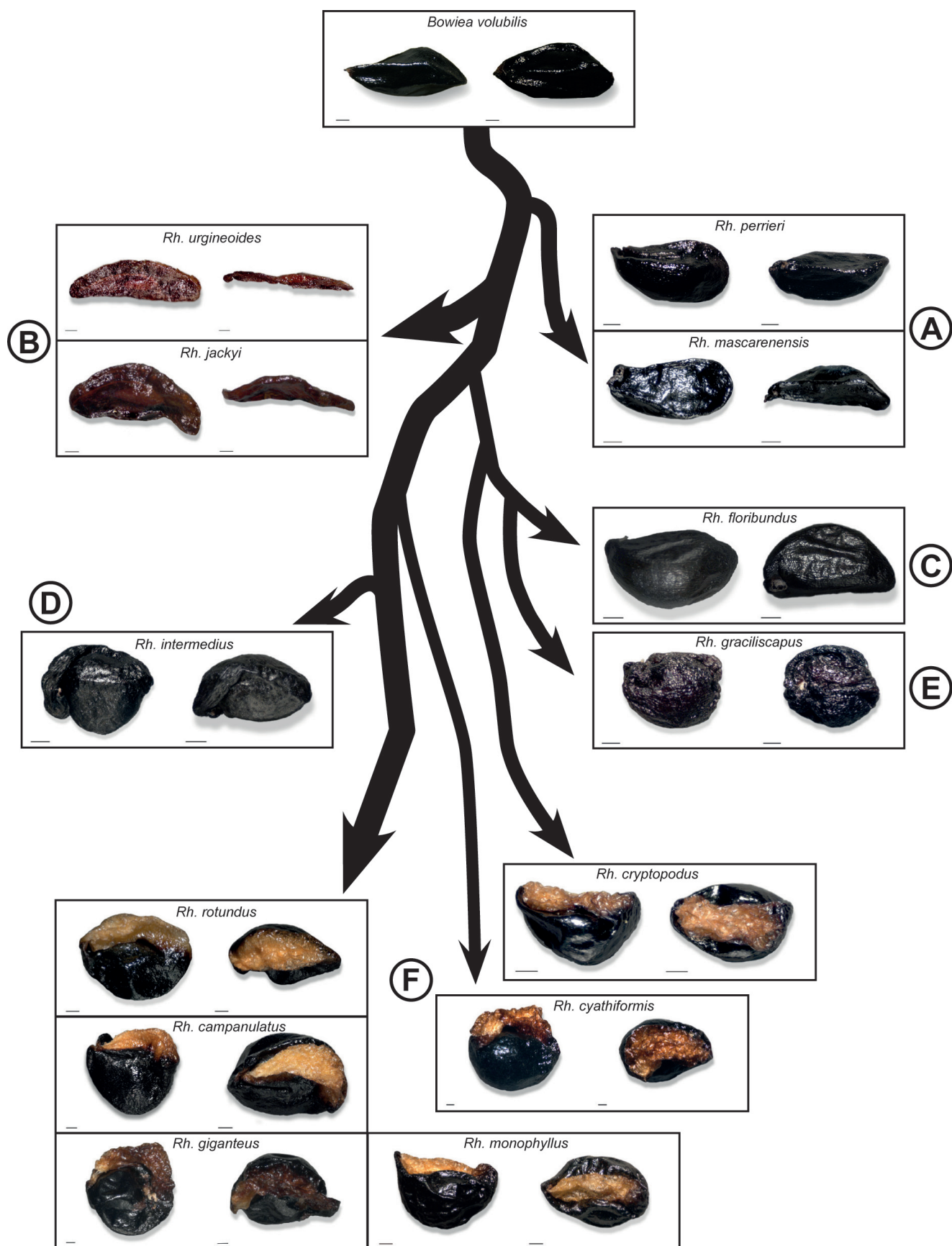


Fig. 1. Possible evolution of *Rhodocodon* seeds according to KNIRSCH 2012. – Scale bars = 0.5 mm.

Endosperm: Oblate, ovate in longitudinal section, circular in cross section, with smooth outline. Length 1.64 mm. Width 1.08 mm. Height 1.04 mm. Area of longitudinal section 1.42 mm². Area of cross section 0.84 mm².

Seed: D-shaped in lateral view, irregularly circular in cross section. Shape approximately corresponding to the shape of the endosperm, defined by endosperm and testa. Thousand-seed weight 1.35 g. Length 2.7 mm. Height 1.6 mm. Width 1.25 mm. Area of lateral view 3.44 mm². Area of longitudinal section 3.44 mm². Area of cross section 1.87 mm². Edges/strips/wings absent. Testa partially tight-fitting on the endosperm, without appendices, black, glossy, cell pattern visible. Raphe and hilum color different than testa.

Seedcoat surface: Raphe elevated. Stomata absent.

Primary sculpture: Cell area 5356.3 µm². Cell perimeter 364.72 µm. Frame width 130.83 µm. Frame height 80.59 µm. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries channeled, cell corners not divergent, periclinal cell wall concave or plane.

Secondary sculpture: Surface of the outer cell wall verrucose, with waxes.

Tertiary sculpture: Waxlayer plane, smooth.

3.3. *Rhodocodon cyathiformis* H. PERRIER ex KNIRSCH, MART.-AZORÍN & WETSCHNIG (Fig. 1F, 3a–f, 9c)

Embryo: Shape of longitudinal section lineal, straight. Length of axis 2.49 mm. Width 0.56 mm. Height 0.46 mm. Area of longitudinal section 0.95 mm². Area of cross section 0.18 mm².

Endosperm: Not oblate, ovate in longitudinal section, ovate in cross section, with crenate outline. Length 3.8 mm. Width 2.49 mm. Height 2.77 mm. Area of longitudinal section 7.98 mm². Area of cross section 5.52 mm².

Seed: Ovate in lateral view, elliptic in cross section. Shape approximately corresponding to the shape of the endosperm, defined by endosperm and testa. Thousand-seed weight 22.53 g. Length 5.96 mm. Height 4.93 mm. Width 3.51 mm. Area of lateral view 23.83 mm². Area of longitudinal section 23.83 mm². Area of cross section 11.33 mm². Edges/strips/wings absent. Testa not connected with the endosperm, without appendices, black, glossy, cell pattern visible. Raphe with different color than testa. Hilum colored like testa.

Seedcoat surface: Raphe elevated. Stomata absent.

Primary sculpture: Cell area 24409 µm². Cell perimeter 1187.3 µm. Frame width 311.11 µm. Frame height 280.62 µm. Cell shape uniform, gener-

ally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, cell corners recessed, periclinal cell wall plane.

Secondary sculpture: Surface of the outer cell wall smooth, with waxes.

Tertiary sculpture: Waxlayer plane, smooth.

3.4. *Rhodocodon floribundus* H. PERRIER ex KNIRSCH, MART.-AZORÍN & WETSCHNIG (Fig. 1C, 3g–l, 9d)

Embryo: Shape of longitudinal section clubbed, curved. Length of axis 1.98 mm. Width 0.24 mm. Height 0.35 mm. Area of longitudinal section 0.48 mm². Area of cross section 0.07 mm².

Endosperm: Oblate, D-shaped in longitudinal section, obtuse-triangular in cross section, with crenate outline. Length 2.48 mm. Width 1.64 mm. Height 1.8 mm. Area of longitudinal section 3.03 mm². Area of cross section 2.07 mm².

Seed: D-shaped in lateral view, obtuse-triangular in cross section. Shape approximately corresponding to the shape of endosperm, defined by endosperm and testa. Thousand-seed weight 3.95 g. Length 3.5 mm. Height 2.08 mm. Width 2.12 mm. Area of lateral view 5.54 mm². Area of longitudinal section 5.54 mm². Area of cross section 2.69 mm². Edges/strips/wings absent. Testa not connected with the endosperm, without appendices, black, glossy, cell pattern visible. Raphe colored like testa. Hilum with different color than testa.

Seedcoat surface: Raphe not visible. Stomata absent.

Primary sculpture: Cell area 13750 µm². Cell perimeter 660.39 µm. Frame width 240.61 µm. Frame height 152.11 µm. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries channeled, cell corners not divergent, periclinal cell wall concave or plane.

Secondary sculpture: Surface of the outer cell wall reticulate, with waxes.

Tertiary sculpture: Waxlayer plane, smooth.

3.5. *Rhodocodon giganteus* ined.
(Fig. 1F, 4a–f, 10)

Embryo: Shape of longitudinal section clubbed, straight. Length of axis 1.73 mm. Width 0.55 mm. Height 0.6 mm. Area of longitudinal section 0.54 mm². Area of cross section 0.28 mm².

Endosperm: Not oblate, circular in longitudinal section, ovate in cross section, with smooth outline. Length 3.35 mm. Width 2.88 mm. Height 3.69 mm. Area of longitudinal section 9.6 mm². Area of cross section 7.23 mm².

Seed: Elliptic in lateral view, irregularly circular in cross section. Shape partially correspond-

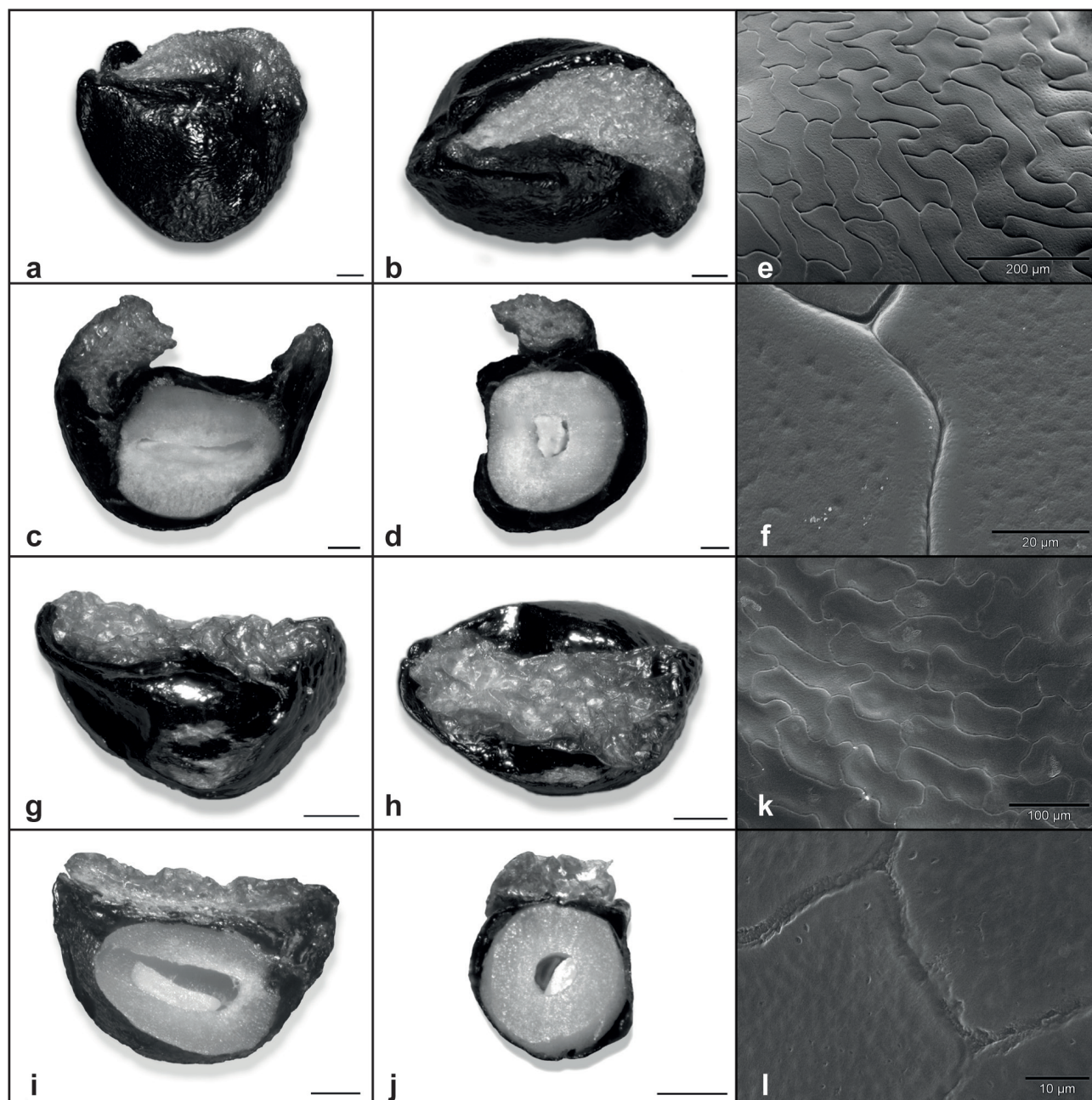


Fig. 2. (a–f) *Rhodocodon campanulatus*, (g–l) *Rhodocodon cryptopodus*. – (a, g) Seed in longitudinal view. – (b, h) Seed in raphal view. – (c, i) Seed in longitudinal section. – (d, j) Seed in cross section. – (e–f), (k–l) Scanning electron micrograph of seed surface. – Scale bars: a–d, g–j = 0.5 mm, e = 200 μ m, f = 20 μ m, k = 100 μ m, l = 10 μ m.

ing to the shape of the endosperm, defined by testa. Thousand-seed weight 26.43 g. Length 5.14 mm. Height 4.32 mm. Width 3.39 mm. Area of lateral view 16.11 mm². Area of longitudinal section 16.11 mm². Edges/strips/wings absent. Testa not connected with the endosperm, without appendices, black, glossy, cell pattern visible. Raphe and hilum color different than testa.

Seedcoat surface: Raphe elevated. Stomata absent.

Primary sculpture: Cell area 18601 μ m². Cell perimeter 927.64 μ m. Frame width 220.28 μ m. Frame height 218.68 μ m. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries channeled, cell corners not divergent, periclinal cell wall convex.

Secondary sculpture: Surface of the outer cell wall reticulate, with waxes.

Tertiary sculpture: Waxlayer plane, smooth.

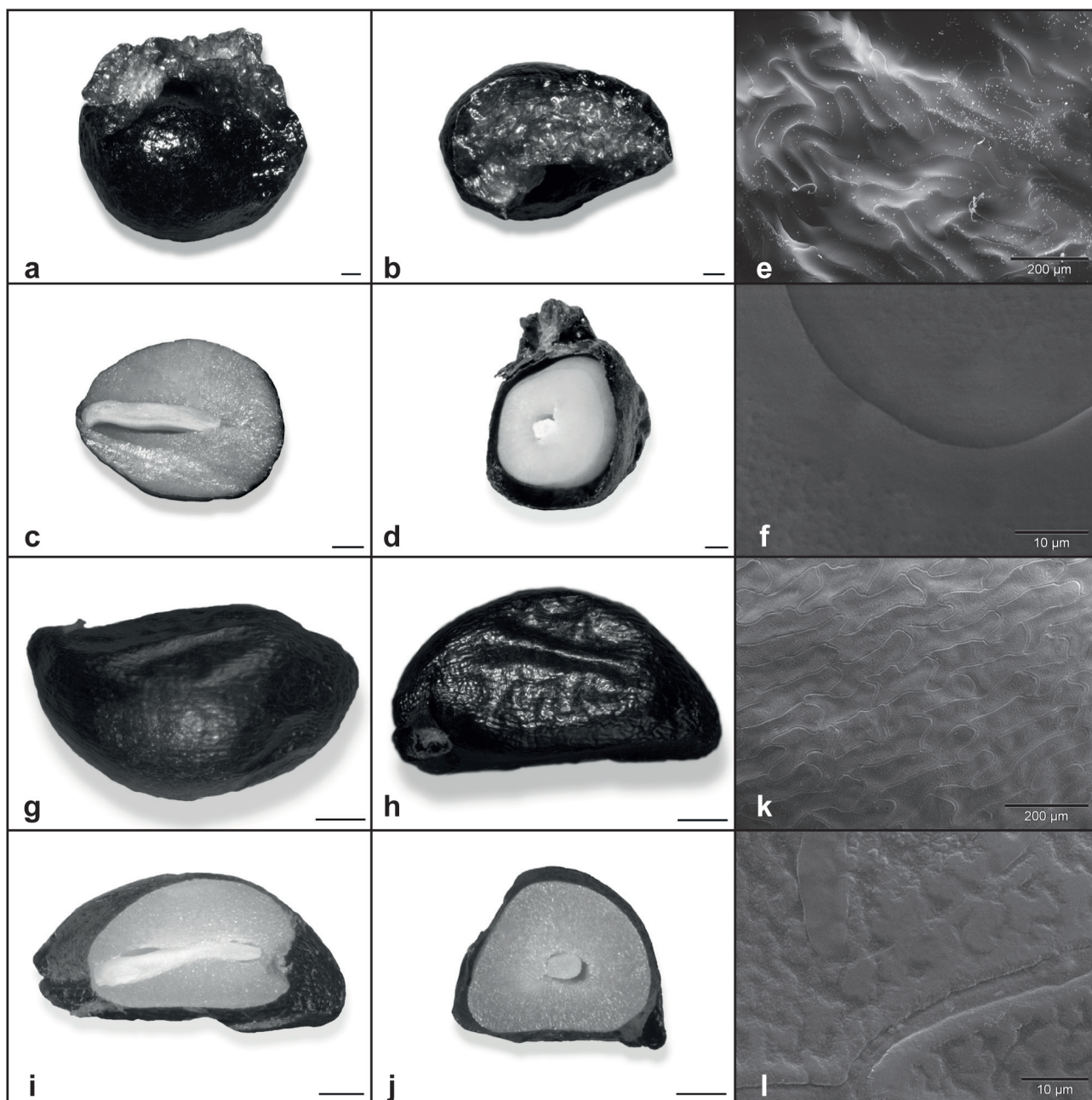


Fig. 3. (a–f) *Rhodocodon cyathiformis*, (g–l) *Rhodocodon floribundus*. – (a, g) Seed in longitudinal view. – (b, h) Seed in raphal view. – (c, i) Seed in longitudinal section. – (d, j) Seed in cross section. – (e–f), (k–l) Scanning electron micrograph of seed surface. – Scale bars: a–d, g–j = 0.5 mm, e = 200 µm, f = 10 µm, k = 200 µm, l = 10 µm.

3.6. *Rhodocodon graciliscapus* KNIRSCH, MART. – AZORÍN & WETSCHNIG (Fig. 1E, 4g–l)

Embryo: Shape of longitudinal section clubbed, curved. Length of axis 1.67 mm. Width 0.3 mm. Height 0.3 mm. Area of longitudinal section 0.44 mm². Area of cross section 0.07 mm².

Endosperm: Not oblate, pear-shaped in longitudinal section, obtuse-triangular in cross section, with crenate outline. Length 2.05 mm. Width

1.65 mm. Height 1.65 mm. Area of longitudinal section 2.53 mm². Area of cross section 1.93 mm².

Seed: D-shaped in lateral view, irregularly circular in cross section. Shape partially corresponding to the shape of the endosperm, defined by endosperm and testa. Thousand-seed weight 2.9 g. Length 2.9 mm. Height 2.4 mm. Width 2.34 mm. Area of lateral view 5.1 mm². Area of longitudinal section 5.1 mm². Area of cross section 4.28 mm². Edges/strips/wings absent. Testa not connected

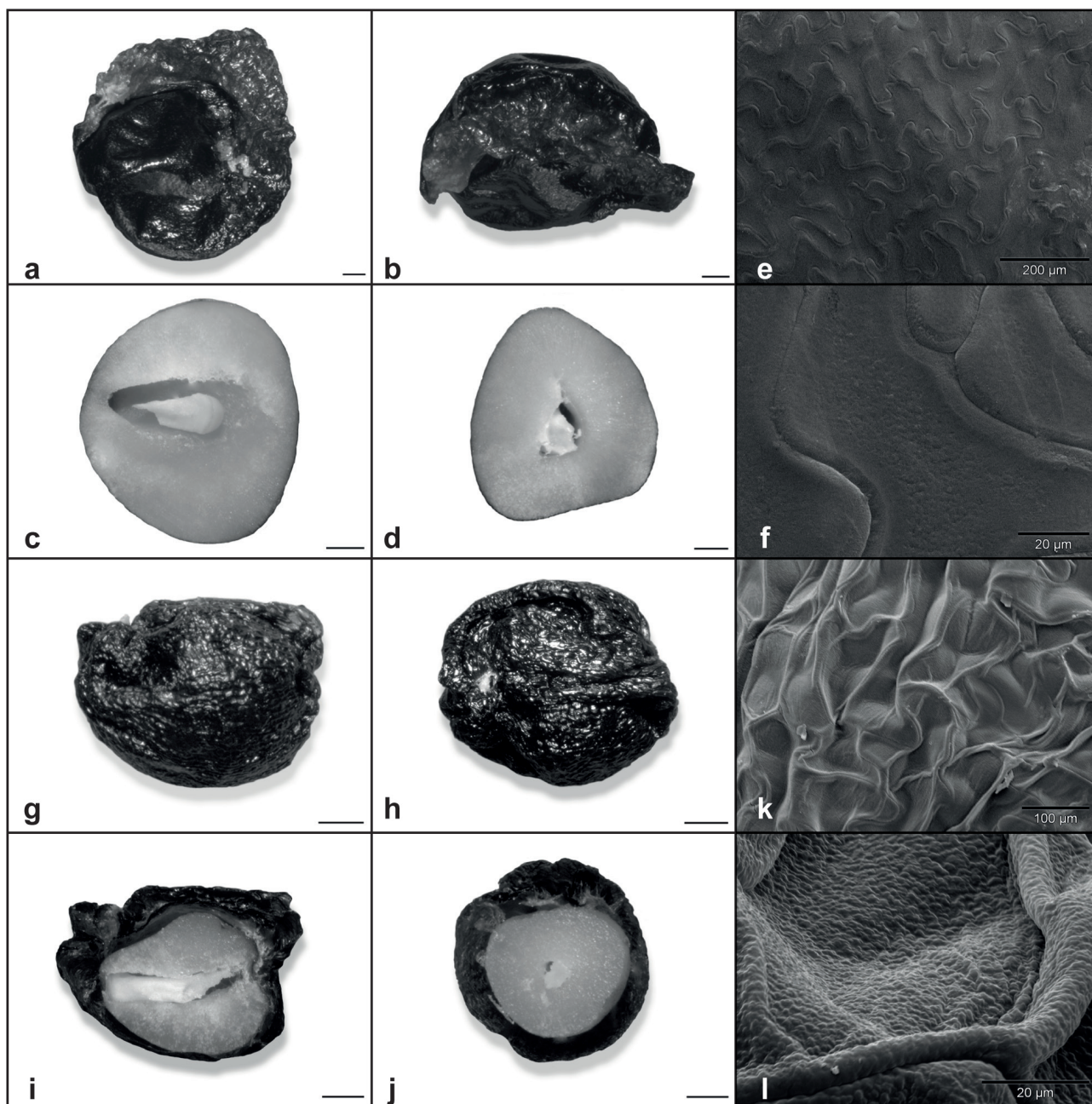


Fig. 4. (a–f) *Rhodocodon giganteus* ined., (g–l) *Rhodocodon graciliscapus*. – (a, g) Seed in longitudinal view. – (b, h) Seed in raphal view. – (c, i) Seed in longitudinal section. – (d, j) Seed in cross section. – (e–f), (k–l) Scanning electron micrograph of seed surface. – Scale bars: a–d, g–j = 0.5 mm, e = 200 µm, f = 20 µm, k = 100 µm, l = 20 µm.

with the endosperm, without appendices, black, glossy, cell pattern visible. Raphe colored like testa. Hilum with different color than testa.

Seedcoat surface: Raphe not visible. Stomata absent.

Primary sculpture: Cell area 10908 µm². Cell perimeter 493.11 µm. Frame width 128.2 µm. Frame height 167.78 µm. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries channeled,

cell corners not divergent, periclinal cell wall concave, collapsed (e.g. BARTHOLOTT & EHLE 1977: 32).

Secondary sculpture: Surface of the outer cell wall verrucose.

3.7. *Rhodocodon intermedius* H. PERRIER ex KNIRSCH, MART.-AZORÍN & WETSCHNIG (Fig. 1D, 5a–f)

Embryo: Shape of longitudinal section cylindrical, straight. Length of axis 0.56 mm. Width 0.41

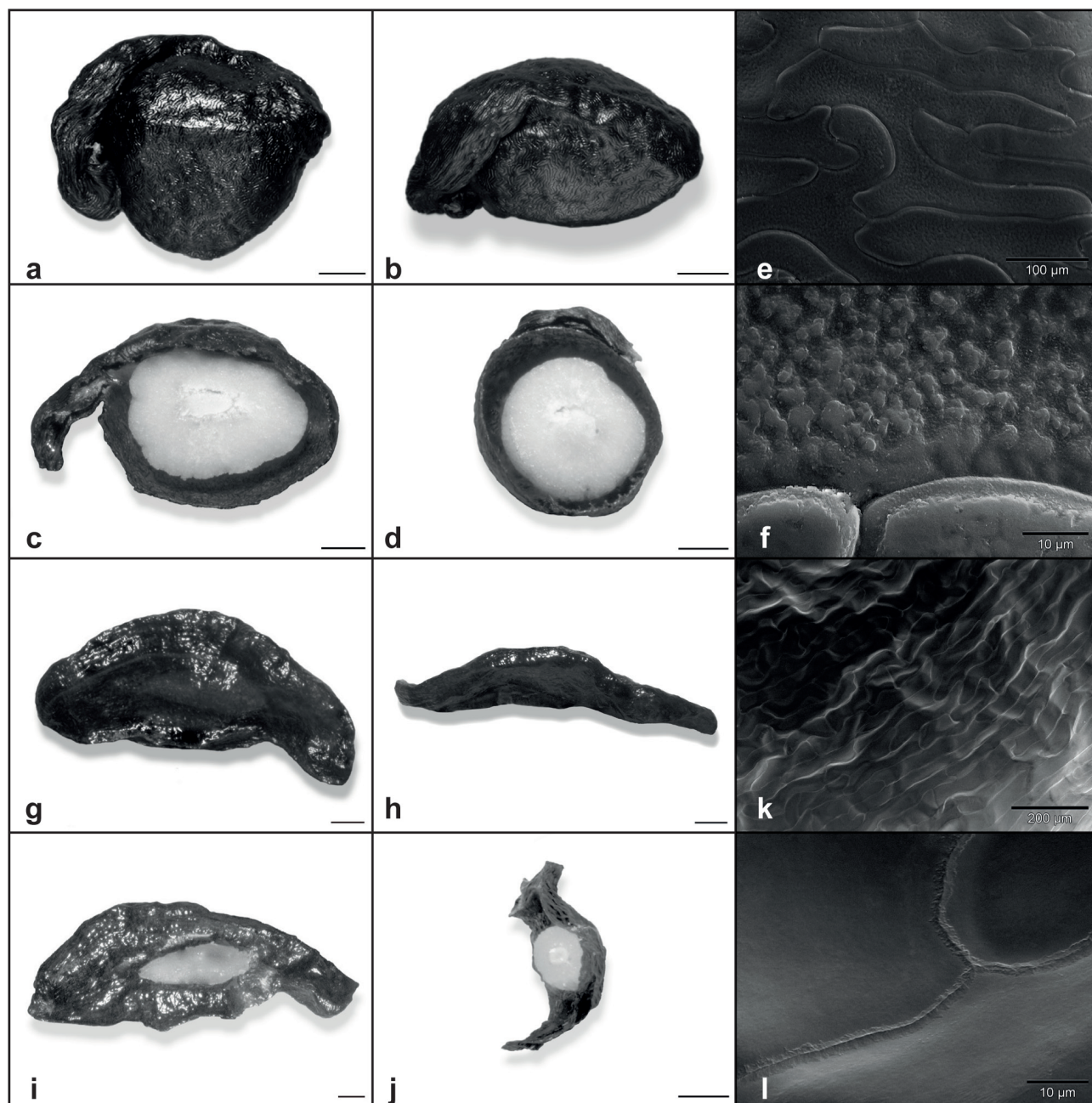


Fig. 5. (a–f) *Rhodocodon intermedius*, (g–l) *Rhodocodon jackyi*. (a, g) – Seed in longitudinal view. – (b, h) Seed in raphal view. – (c, i) Seed in longitudinal section. – (d, j) Seed in cross section. – (e–f), (k–l) Scanning electron micrograph of seed surface. – Scale bars: a–d, g–j = 0.5 mm, e = 100 μ m, f = 10 μ m, k = 200 μ m, l = 10 μ m.

mm. Height 0.25 mm. Area of longitudinal section 0.13 mm². Area of cross section 0.08 mm².

Endosperm: Not oblate, ovate in longitudinal section, circular in cross section, with crenate outline. Length 2 mm. Width 1.4 mm. Height 1.4 mm. Area of longitudinal section 2.22 mm². Area of cross section 1.33 mm².

Seed: D-shaped in lateral view, irregularly circular in cross section. Shape partially corresponding to the shape of the endosperm, defined by en-

dosperm and testa. Thousand-seed weight 2.41 g. Length 3.11 mm. Height 2.3 mm. Width 1.7 mm. Area of lateral view 4.6 mm². Area of longitudinal section 4.6 mm². Area of cross section 2.45 mm². Edges/strips/wings absent. Testa not connected with the endosperm, without appendices, black, glossy, cell pattern visible. Raphe colored like testa. Hilum with different color than testa.

Seedcoat surface: Raphe visible, elevated, with conspicuously elongated cells. Stomata absent.

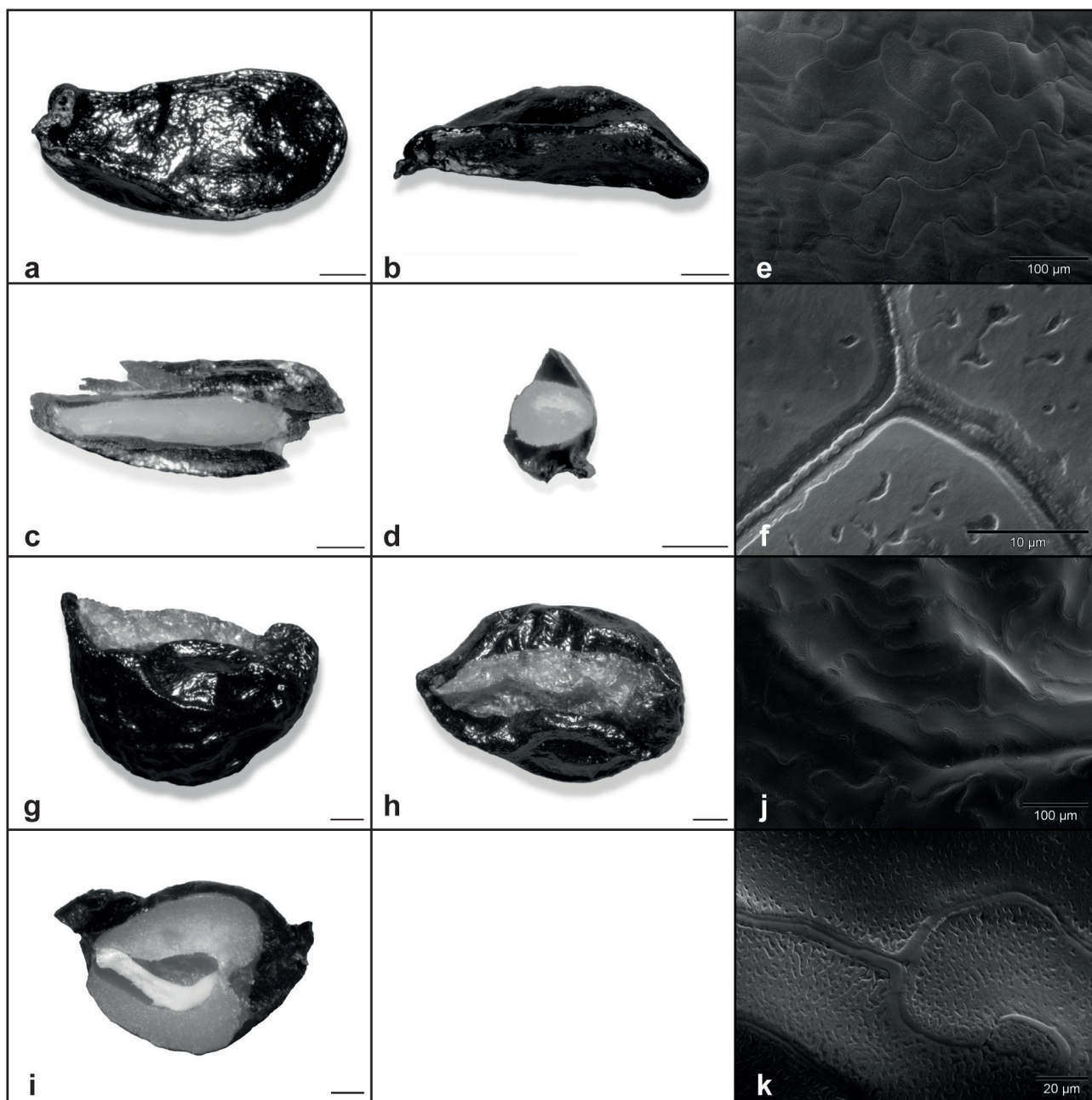


Fig. 6. (a–f) *Rhodocodon mascarenensis*, (g–k) *Rhodocodon monophyllus*. – (a, g) Seed in longitudinal view. – (b, h) Seed in raphal view. – (c, i) Seed in longitudinal section. – (d) Seed in cross section. – (e–f), (j–k) Scanning electron micrograph of seed surface. – Scale bars: a–d, g–i = 0.5 mm, e = 100 µm, f = 10 µm, j = 100 µm, k = 20 µm.

Primary sculpture: Cell area 21076 µm². Cell-perimeter 1133.5 µm. Frame width 370.8 µm. Frame height 190.66 µm. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries channeled, cell corners not divergent, periclinal cell wall plane.

Secondary sculpture: Surface of the outer cell wall verrucose, with waxes.

Tertiary sculpture: Wax layer plane, verrucose.

3.8. *Rhodocodon jackyi* KNIRSCH, MART.-AZORÍN & WETSCHNIG (Fig. 1B, 5g–l, 9a)

Embryo: Shape of longitudinal section clubbed, straight. Length of axis 1.19 mm. Width 0.19 mm. Height 0.28 mm. Area of longitudinal section 0.23 mm². Area of cross section 0.04 mm².

Endosperm: Oblate, acute-ovate in longitudinal section, circular in cross section, with crenate outline. Length 1.88 mm. Width 0.54 mm. Height

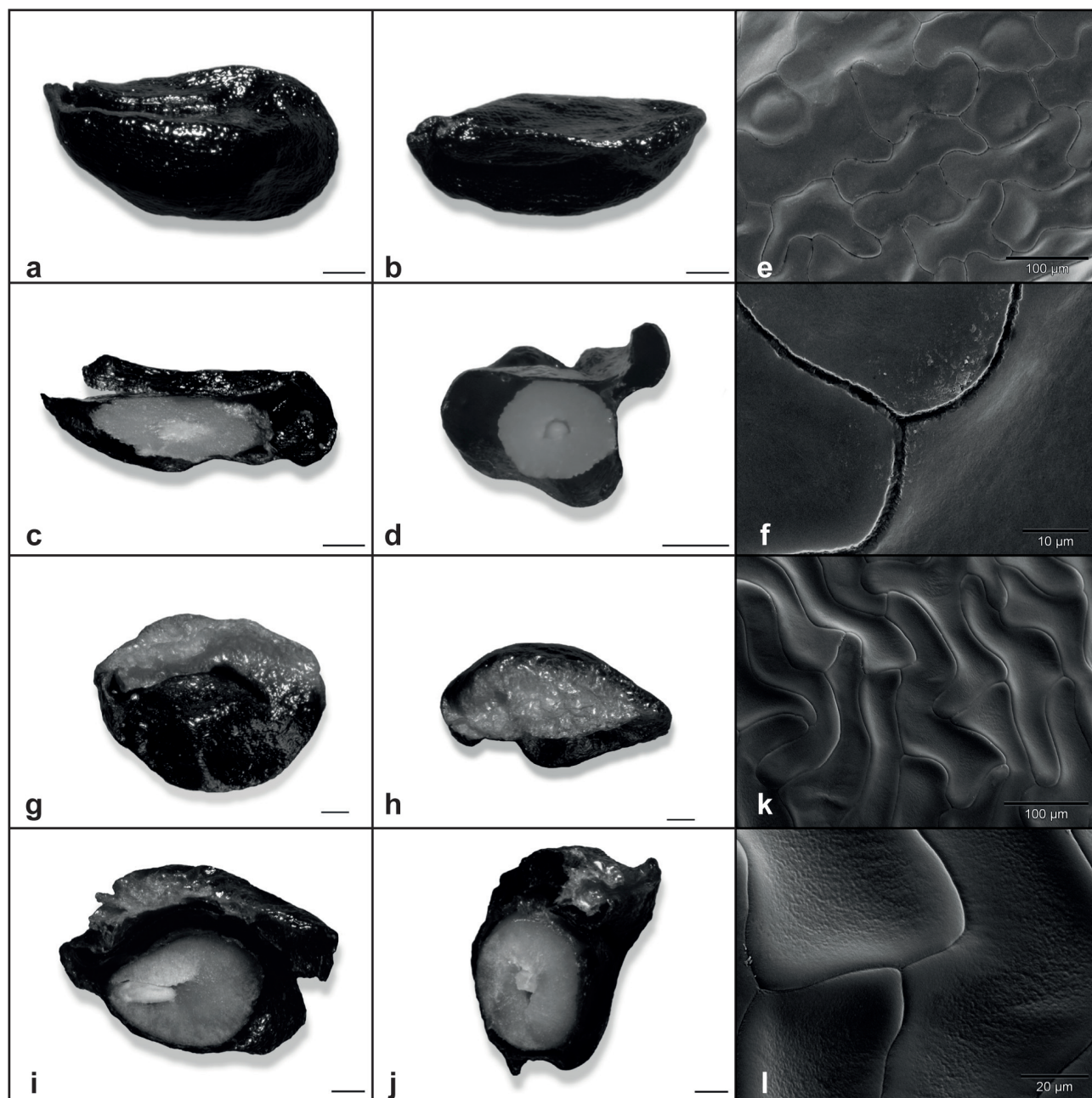


Fig. 7. (a–f) *Rhodocodon perrieri* ined., (g–l) *Rhodocodon rotundus*. – (a, g) Seed in longitudinal view. – (b, h) Seed in raphal view. – (c, i) Seed in longitudinal section. – (d, j) Seed in cross section. – (e–f), (k–l) Scanning electron micrograph of seed surface. – Scale bars: a–d, g–j = 0.5 mm, e = 100 µm, f = 10 µm, k = 100 µm, l = 20 µm.

0.64 mm. Area of longitudinal section 0.95 mm². Area of cross section 0.26 mm².

Seed: Curved-elliptic in lateral view, lineal with one bifid end in cross section. Shape not corresponding to the shape of the endosperm, defined by testa. Thousand-seed weight 0.29 g. Length 5 mm. Height 2 mm. Width 0.5 mm. Area of lateral view 8.98 mm². Area of longitudinal section 8.98 mm². Area of cross section 0.52 mm². Edges/strips/wings present. Testa tight-fitting on en-

dosperm, without appendices, copper brown, glossy, cell pattern visible. Raphe colored like testa. Hilum with different color than testa.

Seedcoat surface: Raphe not visible. Stomata absent.

Primary sculpture: Cell area 8639 µm². Cell perimeter 525.71 µm. Frame width 183.88 µm. Frame height 154.19 µm. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries channeled,

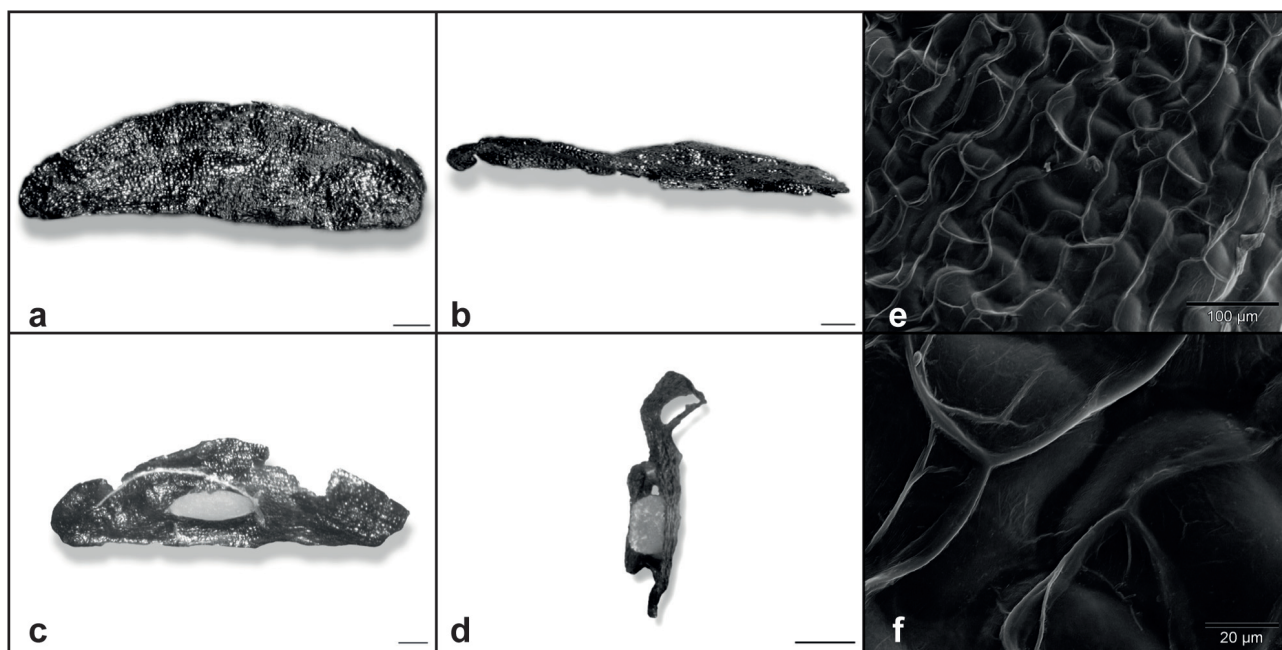


Fig. 8. *Rhodocodon urGINEOIDES*. – (a) Seed in longitudinal view. – (b) Seed in raphal view. – (c) Seed in longitudinal section. – (d) Seed in cross section. – (e, f) Scanning electron micrograph of seed surface. – Scale bars: a–d = 0.5 mm, e = 100 µm, f = 20 µm.

cell corners not divergent, periclinal cell wall concave or plane.

Secondary sculpture: Surface of the outer cell wall smooth, with waxes.

Tertiary sculpture: Wax layer plane, smooth.

3.9. *Rhodocodon mascarensis* (BAKER) KNIRSCH, MART.-AZORÍN & WETSCHNIG (Fig. 1A, 6a–f)

Embryo: Shape of longitudinal section lineal, curved. Length of axis 1.02 mm. Width 0.21 mm. Height 0.23 mm. Area of longitudinal section 0.18 mm². Area of cross section 0.03 mm².

Endosperm: Oblate, fusiform in longitudinal section, circular in cross section, with crenate outline. Length 1.95 mm. Width 0.61 mm. Height 0.6 mm. Area of longitudinal section 0.81 mm². Area of cross section 0.24 mm².

Seed: Lanceolate in lateral view, cross section circular in outline, with multiple edges. Shape partially corresponding to the shape of the endosperm, defined by endosperm and testa. Thousand-seed weight 0.57 g. Length 2.56 mm. Height 0.73 mm. Width 1.27 mm. Area of lateral view 1.37 mm². Area of longitudinal section 1.38 mm². Area of cross section 0.35 mm². Edges/strips/wings present. Testa not connected with the endosperm, without appendices, black, glossy, cell pattern visible. Raphe and hilum colored like testa.

Seedcoat surface: Raphe not visible. Stomata absent.

Primary sculpture: Cell area 5401.6 µm². Cell-perimeter 432.94 µm. Frame width 263.16 µm. Frame height 128.07 µm. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries channeled, cell corners not divergent, periclinal cell wall plane.

Secondary sculpture: Surface of the outer cell wall foveolate, with waxes.

Tertiary sculpture: Wax layer plane, verrucose.

3.10. *Rhodocodon monophyllus* KNIRSCH, MART.-AZORÍN & WETSCHNIG (Fig. 1F, 6g–k)

Embryo: Shape of longitudinal section cylindric, curved. Length of axis 2.16 mm. Height 0.36 mm. Area of longitudinal section 0.66 mm².

Endosperm: Oblate, pear-shaped in longitudinal section, with smooth outline. Length 2.64 mm. Height 2.45 mm. Area of longitudinal section 4.75 mm².

Seed: D-shaped in cross section. Shape not corresponding to the shape of the endosperm, defined by testa. Thousand-seed weight 6.8 g. Length 3.87 mm. Height 2.65 mm. Width 2.63 mm. Area of lateral view 6.5 mm². Area of longitudinal section 6.5 mm². Edges/strips/wings absent. Testa partially tight-fitting on endosperm, without appendices, black, glossy, cell pattern visible. Raphe and hilum color different than testa.

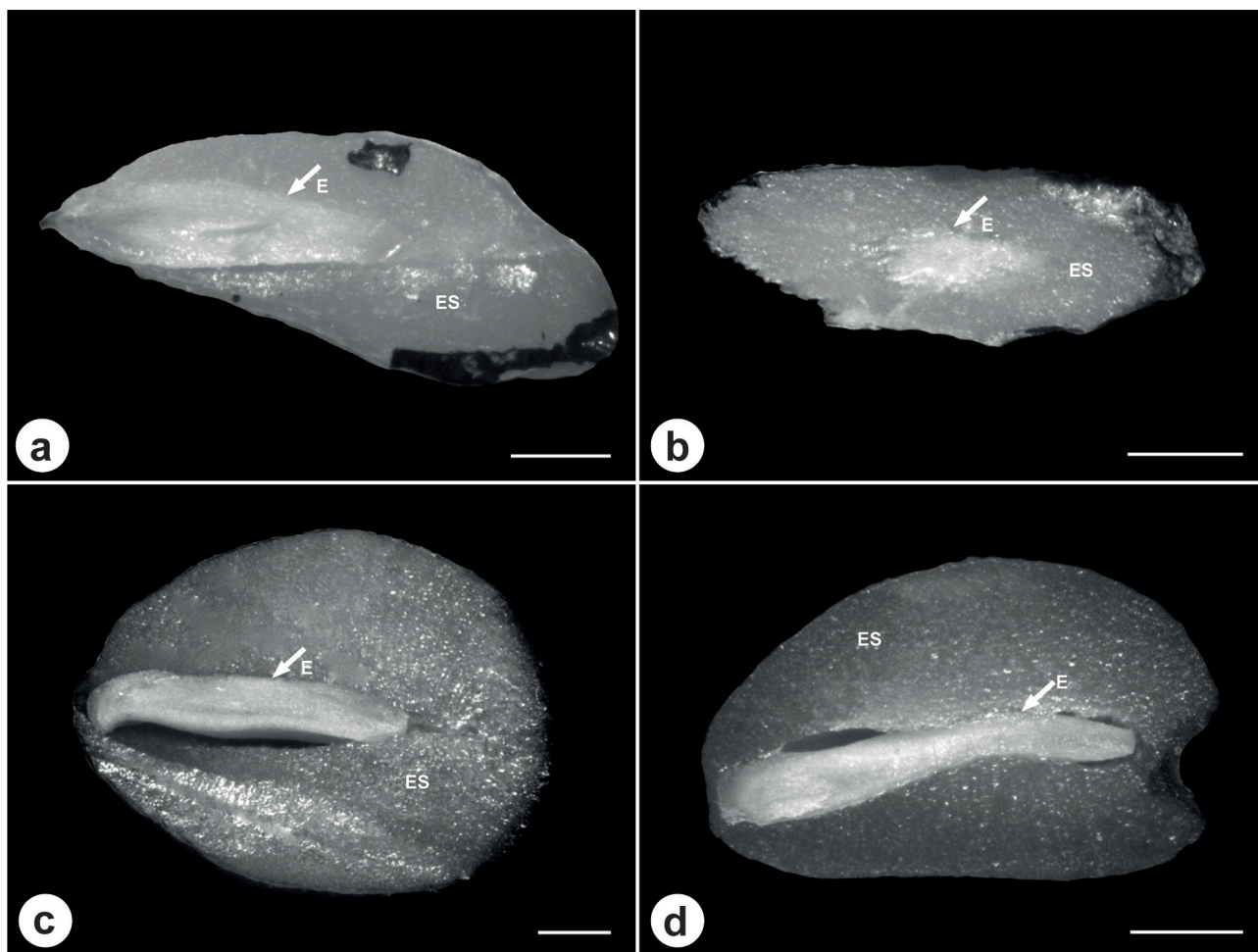


Fig. 9. Light micrographs of longitudinal sections of endosperm (ES) and embryo (E). – (a) *R. jackyi*. – (b) *R. perrieri*. – (c) *R. cyathiformis*. – (d) *R. floribundus*. – Scale bars = 0.5 mm.

Seedcoat surface: Raphe not visible. Stomata absent.

Primary sculpture: Cell area $12570 \mu\text{m}^2$. Cell perimeter $689.53 \mu\text{m}$. Frame width $249.53 \mu\text{m}$. Frame height $116.4 \mu\text{m}$. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries channeled, cell corners not divergent, periclinal cell wall convex or plane.

Secondary sculpture: Surface of the outer cell wall reticulate, with waxes.

Tertiary sculpture: Wax layer plane, smooth.

3.11. *Rhodocodon perrieri* ined. (Fig. 1A, 7a–f, 9b)

Embryo: Shape of longitudinal section cylindrical, straight. Length of axis 0.83 mm . Width 0.18 mm . Height 0.18 mm . Area of longitudinal section 0.14 mm^2 . Area of cross section 0.03 mm^2 .

Endosperm: Oblate, fusiform in longitudinal section, ovate in cross section, with crenate outline.

Length 2.17 mm . Width 0.8 mm . Height 0.8 mm . Area of longitudinal section 1.23 mm^2 . Area of cross section 0.51 mm^2 .

Seed: Drop-shaped in lateral view, often \pm asymmetrical, obtuse-triangular in cross section. Shape partially corresponding to the shape of the endosperm, defined by endosperm and testa. Thousand-seed weight 0.8 g . Length 3.5 mm . Height 1.46 mm . Width 1.5 mm . Area of lateral view 4.9 mm^2 . Area of longitudinal section 4.9 mm^2 . Area of cross section 1.18 mm^2 . Edges/strips/wings present. Testa partially tight-fitting on endosperm, without appendices, black, glossy, cell pattern visible. Raphe colored like testa. Hilum with different color than testa.

Seedcoat surface: Raphe not visible. Stomata absent.

Primary sculpture: Cell area $9208.6 \mu\text{m}^2$. Cell perimeter $461.45 \mu\text{m}$. Frame width $157.78 \mu\text{m}$. Frame height $115.47 \mu\text{m}$. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregu-

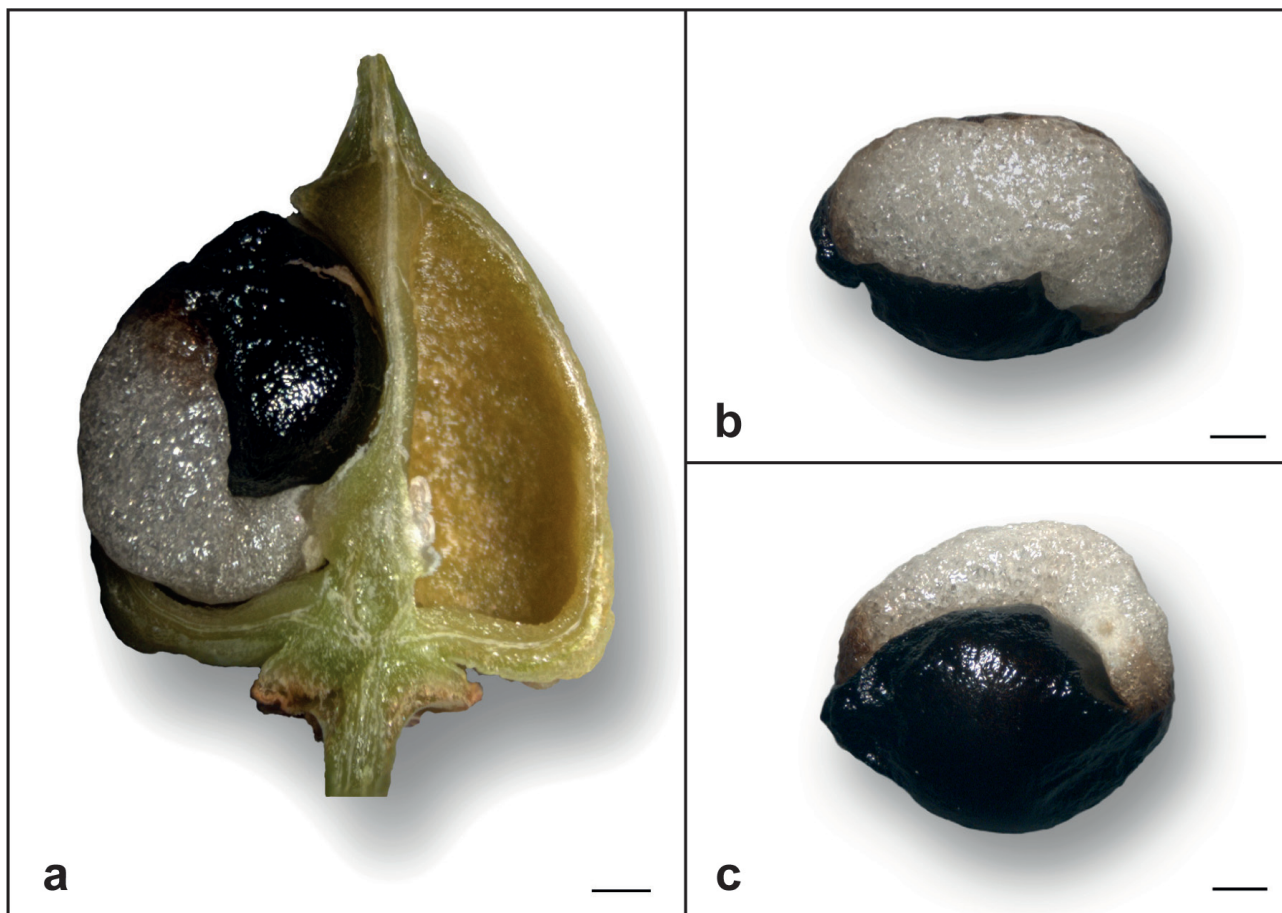


Fig. 10. *Rhodocodon giganteus* ined. – (a) Seed in mature capsule. – (b) Seed in raphal view. – (c) Seed in longitudinal view. – Scale bars = 1 cm.

larly lobate in surface view, boundaries channeled, cell corners not divergent, periclinal cell wall convex or plane.

Secondary sculpture: Surface of the outer cell wall smooth, with waxes.

Tertiary sculpture: Wax layer plane, smooth.

3.12. *Rhodocodon rotundus* H. PERRIER ex KNIRSCH, MART.-AZORÍN & WETSCHNIG (Fig. 1F, 7g–l)

Embryo: Shape of longitudinal section clubbed, straight. Length of axis 0.95 mm. Width 0.32 mm. Height 0.32 mm. Area of longitudinal section 0.26 mm². Area of cross section 0.09 mm².

Endosperm: Not oblate, acute-ovate in longitudinal section, ovate in cross section, with crenate outline. Length 2.34 mm. Width 1.5 mm. Height 1.7 mm. Area of longitudinal section 2.9 mm². Area of cross section 2.36 mm².

Seed: Elliptic in lateral view and in cross section. Shape partially corresponding to the shape of the endosperm, defined by endosperm and testa. Thousand-seed weight 6.13 g. Length 4.1 mm. Height

3.42 mm. Width 2.2 mm. Area of lateral view 9.2 mm². Area of longitudinal section 9.2 mm². Area of cross section 5.06 mm². Edges/strips/wings present. Testa partially tight-fitting on endosperm, without appendices, black, glossy, cell pattern visible. Raphe and hilum color different than testa.

Seedcoat surface: Raphe elevated. Stomata absent.

Primary sculpture: Cell area 16439 µm². Cell-perimeter 821.76 µm. Frame width 146.15 µm. Frame height 265.43 µm. Cell shape uniform, generally elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries channeled, cell corners not divergent, periclinal cell wall concave.

Secondary sculpture: Surface of the outer cell wall verrucose, with waxes.

Tertiary sculpture: Wax layer plane, verrucose.

3.13. *Rhodocodon urGINEOIDES* BAKER (Fig. 1B, 8a–f)

Embryo: Shape of longitudinal section lineal, curved. Length of axis 1.3 mm. Width 0.15 mm.

Height 0.15 mm. Area of longitudinal section 0.13 mm². Area of cross section 0.01 mm².

Endosperm: Oblate, elliptic in longitudinal section, elliptic in cross section, with crenate outline. Length 1.44 mm. Width 0.46 mm. Height 0.49 mm. Area of longitudinal section 0.52 mm². Area of cross section 0.12 mm².

Seed: D-shaped in lateral view, elliptic with two wings in cross section. Shape not corresponding to the shape of the endosperm, defined by testa. Thousand-seed weight 0.21 g. Length 6.1 mm. Height 1.81 mm. Width 0.7 mm. Area of lateral view 8.33 mm². Area of longitudinal section 8.33 mm². Area of cross section 0.5 mm². Edges/strips/wings present. Testa not connected with the endosperm, without appendices, brown, matt, cell pattern visible. Raphe and hilum colored like testa.

Seedcoat surface: Raphe not visible. Stomata absent.

Primary sculpture: Cell area 5747.2 µm². Cell-perimeter 389.07 µm. Frame width 116 µm. Frame height 134.62 µm. Cell shape uniform, generally elongate-tetragonal or elongate-polygonal. Anticlinal cell wall irregularly lobate in surface view, boundaries raised or plane, cell corners not divergent, periclinal cell wall concave.

Secondary sculpture: Surface of the outer cell wall smooth.

3.14. Key to the seeds of investigated species of *Rhodocodon*

1. Surface of the testa (outer cell walls):
 - smooth 2
 - foveolate 4
 - verrucose 5
 - reticulate 8
- 2(1). Shape of embryo in longitudinal section:
 - lineal 3
 - clubbed ***R. jackyi***
 - cylindrical ***R. perrieri***
- 3(2). Embryo straight; endosperm not oblate; testa black, glossy ***R. cyathiformis***
 Embryo curved; endosperm oblate; testa brown, matt ***R. urGINEOIDES***
- 4(1). Embryo lineal in longitudinal section; endosperm oblate; raphe with same color as testa, not visible ***R. mascarenensis***
 Embryo cylindrical in longitudinal section; endosperm not oblate; raphe with different color than testa, elevated ***R. campanulatus***
- 5(1). Embryo clubbed in longitudinal section 6
 Embryo cylindrical in longitudinal section .. 7
- 6(5). Embryo straight; raphe with different color than testa, elevated; periclinal cell wall concave ***R. rotundus***

- Embryo curved; raphe not visible; periclinal cell wall collapsed ***R. graciliscapus***
- 7(5). Endosperm not oblate; raphe with same color as testa ***R. intermedius***
 Endosperm oblate; raphe with different color than testa ***R. cryptopodus***
- 8(1). Embryo clubbed in longitudinal section; periclinal cell wall concave 9
 Embryo cylindrical in longitudinal section; periclinal cell wall convex or plane
 ***R. monophyllus***
- 9(8). Embryo straight; endosperm not oblate; raphe with different color than testa, elevated
 ***R. giganteus***
 Embryo curved; endosperm oblate; raphe with same color as testa, not visible
 ***R. floribundus***

4. Discussion

Seed morphology in *Urgineoideae* is relatively homogeneous, in most cases the seeds are light, flattened and winged with a loose testa, adapted to wind dispersal.

In contrast, our study reveals a great variability in seed morphology in *Rhodocodon* that indicates adaptations to different dispersal methods, ranging from wind dispersal to zoochory (probably myrmecochory), based on the presence of a distinct raphe in some species, although the latter needs confirmation.

Bowiea volubilis HARVEY ex HOOKER (1867: t. 5619) is considered basal in the phylogenetic studies of *Urgineoideae* (PFOSSER & SPETA 1999) and it shows depressed and subellipsoid, relatively heavy seeds (Fig. 1), a rare character in this subfamily.

Fig. 1A–F ('groups A–F') shows a possible evolution of *Rhodocodon* seeds, according to the phylogenetic studies of KNIRSCH 2012, with *R. mascarenensis* (Fig. 1A, Fig. 6a–f) as the basal representative within the genus. Although there are no genetic data available for *R. perrieri* to support a close relationship to *R. mascarenensis*, the morphological data suggested to unite them in group A. The seeds are depressed, with a glossy, dark colored testa, which is more or less unconnected to the fusiform endosperm, approximately like in *Bowiea*. *Rhodocodon perrieri* (Fig. 1A, Fig. 7a–f) differs by its cylindrical embryo (Fig. 7c) and smooth outer cell wall surface.

Group B represents modified species with very light, flat and winged seeds (HUBER 1969). *Rhodocodon urGINEOIDES* (Fig. 1B, Fig. 8a–f) occurs mostly epiphytic in moss on trees (or rocks) (KNIRSCH & al. 2015). The seeds are absolute lightweights (thousand-seed weight: 0.21 g), well adapted to wind dispersal. The endosperm is tiny in relation to the brown, wing-like testa, which consists only of a few

cell layers. If we consider that the second basal clade in *Urgineoideae*, the genus *Rhadamanthus* s.str., shows a similar type of light-weight seeds, this might even support another hypothesis for the evolution of the group: small, light seeds could be interpreted as an ancestral character in *Rhodocodon*, from which the other types evolved secondarily. This theory would also be in line with a presumed wind dispersal of ancestral species from Africa to Madagascar. Seed morphology of *R. jackyi* (Fig. 1B, Fig. 5g–l) is very similar to *R. urGINEOIDES*. Both species are distinguished by the shape of their embryos. *R. urGINEOIDES* shows a lineal shape, *R. jackyi* a clubbed one (Fig. 9a).

Rhodocodon floribundus (Fig. 1C) and *R. graciliscapus* (Fig. 1E) appear to be somewhat isolated. Especially *R. graciliscapus* differs by its collapsed periclinal cell wall (Fig. 4k) which finds its expression in a “wrinkly” testa (Fig. 4g). From the seed morphological point of view, their close relationship is surprising, in contrast to the phylogenetic studies of KNIRSCH 2012. Similarities can be found in their endosperm characteristics.

The seeds of group F look similar with their prominent saffron-colored raphe and glossy testa, but these characteristics are not useful for species delimitation or taxonomic boundaries. However, they differ considerably in their cell wall and embryo characteristics. Preliminary phylogenetic work by KNIRSCH 2012 suggests that *R. cryptopodus* (Fig. 1F, Fig. 2g–l) is closely related to both *R. floribundus* (Fig. 1C, Fig. 3g–l) and *R. graciliscapus* (Fig. 1E, Fig. 4g–l), despite their different seed morphology, though this relationship needs confirmation.

The yellow raphe is a very interesting character. Fig. 10a–c shows a nearly ripe seed with a foam-like white raphe, which turns into a yellow parchment-like structure within a few minutes (Fig. 1F). It seems that the raphe could be an intermediary storage tissue or simply a spacer in the capsule. Currently nothing is known about the chemistry of this structure. The possible function of the raphe could be related with zoochory, but further research is necessary. Contrary to the yellow raphe in group F, the black prominent raphe of *R. intermedius* (Fig. 1D, Fig. 5a–f) is suitable for determination and a unique trait within the subfamily *Urgineoideae*.

All members of groups C–F grow in similar habitats. Plants and their seeds can be found on dry, gravelly or sandy soil, mostly near riverbanks. The robust seeds, especially the ones with prominent raphes, could also be adapted to float in water or on the soil surface.

In *Rhodocodon*, seed morphology is very diverse and certainly suitable for species delimitation and determination (see the key to the investigated species in section 3.14. above).

Useful parameters are found in the nature of cell walls, especially their surfaces. Four different types can be distinguished: smooth (Fig. 5l), foveolate (BRUDERMANN & al. in prep.) (Fig. 6f), verrucose (Fig. 7l), and reticulate (Fig. 6k).

To this point, we can distinguish three types of embryo shapes in *Hyacinthaceae*: linear (Fig. 9c, 9d), clubbed (Fig. 9a), and cylindric (Fig. 9b). All of these occur also in *Rhodocodon*, but both characters, surface and embryo shape, cannot be assigned to any group (Fig. 1A–F) in an unambiguous way. They are distributed indiscriminately among the 13 species.

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